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(54) Title: HIGH-AFFINITY OLIGONUCLEOTIDE LIGANDS TO IMMUNOGLOBULIN E (IgE)

(57) Abstract

This invention discloses high-affinity oligonucleotide ligands to human Immunoglobulin E (IgE), specifically RNA and ssDNA ligands having the ability to bind to IgE, and the methods for obtaining such ligands. The ligands are capable of inhibiting the interaction of IgE with its receptor.

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HIGH-AFFINITY OLIGONUCLEOTIDE LIGANDS TO IMMUNOGLOBULIN E (IgE)

FIELD OF THE INVENTION

Described herein are methods for identifying and preparing high-affinity nucleic acid ligands to Immunoglobulin E (IgE). The method utilized herein for identifying such nucleic acid ligands is called SELEX, an acronym for Systematic Evolution of Ligands by EXponential enrichment. Specifically disclosed herein are high-affinity nucleic acid ligands. The invention includes high-affinity RNA ligands which bind to IgE and inhibit its ability to interact with the IgE FcεRI receptor. The invention also includes single stranded DNA ligands which bind to IgE and inhibit its ability to interact with the IgE FcεRI receptor.

BACKGROUND OF THE INVENTION

Stimulation of mast cells and basophils upon contact of allergy-specific IgE antibodies with antigens, called immediate hypersensitivity, is one of the most powerful effector mechanisms of the mammalian immune system. Due to a combination of genetic predisposition and environmental stimuli, approximately 20% of the US population is prone to develop an abnormally strong immediate hypersensitivity, a condition known as allergy. Physiological symptoms include increased vascular permeability, vasodilation, smooth muscle contraction, and local inflammation. These and other IgE dependent reactions can cause allergic diseases like allergic rhinitis (hay fever),

asthma, atopic dermatitis (chronic skin irritations) and in the most severe cases can lead to anaphylactic shock, causing death of the individual by asphyxiation and cardiovascular collapse. Common environmental allergens are pollen, dust mites, certain foods, animal dander, fungal spores, and insect venoms.

The first exposure to a specific antigen can lead to the sensitization of the individual. The allergen binds with low specificity to pre-existing IgE in the plasma. This complex interacts with the low affinity receptor FcεRII on antigen presenting cells (APC). The antigen is internalized, proteolytically processed and transported to the surface of the APC by class II MHC molecules. Fragments of the antigen are thereby presented to $CD4^{+}$ T helper cells which in turn activate IgE committed B cells to produce antigen-specific IgE. Normally IgE occurs in the human plasma at a concentration of about 0.2 μg/ml but in atopic patients this level can rise to a concentration of over 10 μg/ml.

Re-exposure to the allergen results in tight binding to the allergen-specific IgE present on the high-affinity receptor FcεRI on the surface of mast cells. Multivalent allergens cause the crosslinking of several receptors in the cell membrane. This triggers an intracellular signaling cascade, leading ultimately to the release of preformed mediators from cytoplasmic granules and the secretion of newly synthesized mediators. These mediators, notably histamines, leukotrienes, prostaglandins, and proteases, in turn cause the wide spectrum of symptoms of the allergic

response. Furthermore, the release of chemotactic cytokines from the mast cell attracts and activates inflammatory cells to the location of antigen exposure. Finally, the release of IL-4 activates B cells to produce more antigen-specific IgE, thereby amplifying the allergic response. For a review, see Sutton *et al.* (1993) Nature 366:421-428.

Mounting evidence indicates that the IgE system has evolved to cope primarily with infections by parasitic worms like *Schistosoma mansoni.* In the absence of such parasites, IgE mediated responses seem to be dispensable and frequently lead to pathologic consequences. Supporting this hypothesis is the fact that murine strains deficient in IgE or the IgE high-affinity receptor (Dombrowicz, *et al.* (1993) Cell 75:969-976) lack the anaphylactic response, but appear otherwise normal.

IgE is a 190 K_D antibody consisting of two ε heavy chains (70 K_D) and two light chains (25 K_D). The heavy chains contain one variable domain (V_H) and four constant domains (C_H1 to C_H4). The light chains contain one variable domain (V_L) and one constant domain (C_L). Each of these immunoglobulin domains consists of about 100 residues and is stabilized by intramolecular sulfur bridges. The heavy and light chains are connected by intermolecular sulfur bridges.

The IgE molecule can be subdivided into the F_{AB} (antigen binding) region, containing the variable and the first constant domains and the F_C (crystalline) region, consisting of the remaining constant domains. The antigen binds to hypervariable sites within the

variable region, whereas the IgE receptors bind to the F_C region. The high-affinity IgE receptor FcεRI contacts a dodeca-peptide sequence located at the N-terminus of the C_H3 domain and the low affinity IgE receptor FcεRII binds to the middle portion of the same domain (reviewed in Sutton, *et al.*, *supra*).

The IgE molecule is significantly bent, reducing its predicted length from 17.5 nm for a planar molecule to 7 nm. This bend occludes one of the two potential FcεRI receptor binding sites resulting in a monovalent IgE molecule which, in the absence of a multivalent allergen, cannot crosslink receptor molecules to initiate the allergic response.

To allow the antigen mediated triggering of the allergic response, IgE must form a complex with the high affinity receptor, FcεRI. FcεRI consists of four transmembrane polypeptides: α, β, and γ_2. The α subunit, FcεRI(α), contains two extracellular immunoglobulin domains and it is the second domain, α(2), that binds to the convex site of the IgE molecule. The dissociation constant of this interaction is approximately 10^{-10} M (Sutton, *et al.*, *supra*). The β and γ chains of FcεRI are necessary to anchor the receptor in the cell membrane, to allow receptor crosslinking, and for signal transduction to initiate the release of mediators from mast cells.

To inhibit immediate hypersensitivity numerous steps of the pathway can be targeted. It should be possible to prevent the synthesis of IgE by binding to and blocking the action of IL-4 or the IL-4 receptor, to prevent mast cell and basophil stimulation by

blocking IgE or the FcεRI IgE receptor, to prevent release of mediators by blocking a step of the intracellular signaling pathway, or to prevent physiological responses of patients by blocking the released mediators. This work demonstrates the use of high-affinity oligonucleotides to human IgE to inhibit the interaction of IgE with the FcεRI receptor.

A method for the *in vitro* evolution of nucleic acid molecules with highly specific binding to target molecules has been developed. This method, Systematic Evolution of Ligands by EXponential enrichment, termed SELEX, is described in United States Patent Application Serial No. 07/536,428, entitled "Systematic Evolution of Ligands by Exponential Enrichment," now abandoned, United States Patent Application Serial No. 07/714,131, filed June 10, 1991, entitled "Nucleic Acid Ligands," United States Patent Application Serial No. 07/931,473, filed August 17, 1992, entitled "Nucleic Acid Ligands," now United States Patent No. 5,270,163 (see also PCT/US91/04078), each of which is herein specifically incorporated by reference. Each of these applications, collectively referred to herein as the SELEX Patent Applications, describes a fundamentally novel method for making a nucleic acid ligand to any desired target molecule.

The SELEX method involves selection from a mixture of candidate oligonucleotides and step-wise iterations of binding, partitioning and amplification, using the same general selection scheme, to achieve virtually any desired criterion of binding affinity and selectivity. Starting from a mixture of nucleic acids, preferably

comprising a segment of randomized sequence, the SELEX method includes steps of contacting the mixture with the target under conditions favorable for binding, partitioning unbound nucleic acids from those nucleic acids which have bound specifically to target molecules, dissociating the nucleic acid-target complexes, amplifying the nucleic acids dissociated from the nucleic acid-target complexes to yield a ligand-enriched mixture of nucleic acids, then reiterating the steps of binding, partitioning, dissociating and amplifying through as many cycles as desired to yield highly specific, high affinity nucleic acid ligands to the target molecule.

The basic SELEX method has been modified to achieve a number of specific objectives. For example, United States Patent Application Serial No. 07/960,093, filed October 14, 1992, entitled "Method for Selecting Nucleic Acids on the Basis of Structure," describes the use of SELEX in conjunction with gel electrophoresis to select nucleic acid molecules with specific structural characteristics, such as bent DNA. United States Patent Application Serial No. 08/123,935, filed September 17, 1993, entitled "Photoselection of Nucleic Acid Ligands" describes a SELEX based method for selecting nucleic acid ligands containing photoreactive groups capable of binding and/or photocrosslinking to and/or photoinactivating a target molecule. United States Patent Application Serial No. 08/134,028, filed October 7, 1993, entitled "High-Affinity Nucleic Acid Ligands That Discriminate Between Theophylline and Caffeine," describes a method for identifying highly

specific nucleic acid ligands able to discriminate between closely related molecules, termed Counter-SELEX. United States Patent Application Serial No. 08/143,564, filed October 25, 1993, entitled "Systematic Evolution of Ligands by EXponential Enrichment: Solution SELEX," describes a SELEX-based method which achieves highly efficient partitioning between oligonucleotides having high and low affinity for a target molecule. United States Patent Application Serial No. 07/964,624, filed October 21, 1992, entitled "Methods of Producing Nucleic Acid Ligands" describes methods for obtaining improved nucleic acid ligands after SELEX has been performed. United States Patent Application Serial No. 08/400,440, filed March 8, 1995, entitled "Systematic Evolution of Ligands by EXponential Enrichment: Chemi-SELEX," describes methods for covalently linking a ligand to its target.

The SELEX method encompasses the identification of high-affinity nucleic acid ligands containing modified nucleotides conferring improved characteristics on the ligand, such as improved *in vivo* stability or improved delivery characteristics. Examples of such modifications include chemical substitutions at the ribose and/or phosphate and/or base positions. SELEX-identified nucleic acid ligands containing modified nucleotides are described in United States Patent Application Serial No. 08/117,991, filed September 8, 1993, entitled "High Affinity Nucleic Acid Ligands Containing Modified Nucleotides," that describes oligonucleotides containing nucleotide

derivatives chemically modified at the 5- and 2'-positions of pyrimidines. United States Patent Application Serial No. 08/134,028, *supra*, describes highly specific nucleic acid ligands containing one or more nucleotides modified with 2'-amino (2'-NH_2), 2'-fluoro (2'-F), and/or 2'-O-methyl (2'-OMe). United States Patent Application Serial No. 08/264,029, filed June 22, 1994, entitled "Novel Method of Preparation of 2' Modified Pyrimidine Intramolecular Nucleophilic Displacement," describes oligonucleotides containing various 2'-modified pyrimidines.

The SELEX method encompasses combining selected oligonucleotides with other selected oligonucleotides and non-oligonucleotide functional units as described in United States Patent Application Serial No. 08/284,063, filed August 2, 1994, entitled "Systematic Evolution of Ligands by Exponential Enrichment: Chimeric SELEX" and United States Patent Application Serial No. 08/234,997, filed April 28, 1994, entitled "Systematic Evolution of Ligands by Exponential Enrichment: Blended SELEX," respectively. These applications allow the combination of the broad array of shapes and other properties, and the efficient amplification and replication properties, of oligonucleotides with the desirable properties of other molecules. Each of the above described patent applications which describe modifications of the basic SELEX procedure are specifically incorporated by reference herein in their entirety.

BRIEF SUMMARY OF THE INVENTION

The present invention includes methods of identifying and producing nucleic acid ligands to Immunoglobulin E (IgE) and the nucleic acid ligands so identified and produced. Nucleic acid sequences are provided that are ligands to IgE. More particularly, RNA and ssDNA sequences are provided that are capable of binding specifically to IgE. The RNA sequences include 2'-NH_2 modified pyrimidines.

Also included in this invention are RNA and ssDNA ligands of IgE that are inhibitors of IgE receptor binding. Specifically, RNA and ssDNA ligands are identified and described which inhibit the interaction of IgE with the FcεRI IgE receptor and thereby inhibit the allergic response elicited by IgE.

Further included in this invention is a method of identifying nucleic acid ligands and nucleic acid ligand sequences to IgE comprising the steps of (a) preparing a candidate mixture of nucleic acids, (b) contacting the candidate mixture of nucleic acid ligands with IgE, (c) partitioning between members of said candidate mixture on the basis of affinity to IgE, and (d) amplifying the selected molecules to yield a mixture of nucleic acids enriched for nucleic acid sequences with a relatively higher affinity for binding to IgE.

More specifically, the present invention includes the RNA ligands to IgE identified according to the above-described method, including those ligands listed in Table 1 (SEQ ID NOS:7-41 and 43-48). Also included are RNA ligands to IgE that are substantially

homologous to any of the given ligands and that have substantially the same ability to bind IgE and inhibit IgE receptor binding. Further included in this invention are RNA ligands to IgE that have substantially the same structural form as the ligands presented herein and that have substantially the same ability to bind IgE and inhibit IgE receptor binding.

Additionally, the present invention includes the ssDNA ligands to IgE identified according to the above-described method, including those ligands listed in Tables 5 and 6 (SEQ ID NOS:51-111). Also included are ssDNA ligands to IgE that are substantially homologous to any of the given ligands and that have substantially the same ability to bind IgE and inhibit IgE receptor binding. Further included in this invention are ssDNA ligands to IgE that have substantially the same structural form as the ligands presented herein and that have substantially the same ability to bind IgE and inhibit IgE receptor binding.

The present invention also includes modified nucleotide sequences based on the RNA and ssDNA ligands identified herein and mixtures of the same.

DETAILED DESCRIPTION OF THE INVENTION

This application describes high-affinity nucleic acid ligands to IgE identified through the method known as SELEX. SELEX is described in U.S. Patent Application Serial No. 07/536,428, entitled Systematic Evolution of Ligands by EXponential Enrichment, now abandoned, U.S. Patent Application Serial No. 07/714,131, filed June 10, 1991, entitled Nucleic Acid

Ligands, United States Patent Application Serial No. 07/931,473, filed August 17, 1992, entitled Nucleic Acid Ligands, now United States Patent No. 5,270,163, (see also PCT/US91/04078). These applications, each specifically incorporated herein by reference, are collectively called the SELEX Patent Applications.

In its most basic form, the SELEX process may be defined by the following series of steps:

1) A candidate mixture of nucleic acids of differing sequence is prepared. The candidate mixture generally includes regions of fixed sequences (i.e., each of the members of the candidate mixture contains the same sequences in the same location) and regions of randomized sequences. The fixed sequence regions are selected either: (a) to assist in the amplification steps described below, (b) to mimic a sequence known to bind to the target, or (c) to enhance the concentration of a given structural arrangement of the nucleic acids in the candidate mixture. The randomized sequences can be totally randomized (*i.e.*, the probability of finding a base at any position being one in four) or only partially randomized (*e.g.*, the probability of finding a base at any location can be selected at any level between 0 and 100 percent).

2) The candidate mixture is contacted with the selected target under conditions favorable for binding between the target and members of the candidate mixture. Under these circumstances, the interaction between the target and the nucleic acids of the candidate mixture can be considered as forming nucleic

acid-target pairs between the target and those nucleic acids having the strongest affinity for the target.

3) The nucleic acids with the highest affinity for the target are partitioned from those nucleic acids with lesser affinity to the target. Because only an extremely small number of sequences (and possibly only one molecule of nucleic acid) corresponding to the highest affinity nucleic acids exist in the candidate mixture, it is generally desirable to set the partitioning criteria so that a significant amount of the nucleic acids in the candidate mixture (approximately 5-50%) are retained during partitioning.

4) Those nucleic acids selected during partitioning as having the relatively higher affinity to the target are then amplified to create a new candidate mixture that is enriched in nucleic acids having a relatively higher affinity for the target.

5) By repeating the partitioning and amplifying steps above, the newly formed candidate mixture contains fewer and fewer unique sequences, and the average degree of affinity of the nucleic acids to the target will generally increase. Taken to its extreme, the SELEX process will yield a candidate mixture containing one or a small number of unique nucleic acids representing those nucleic acids from the original candidate mixture having the highest affinity to the target molecule.

The SELEX Patent Applications describe and elaborate on this process in great detail. Included are targets that can be used in the process; methods for partitioning nucleic acids within a candidate

mixture; and methods for amplifying partitioned nucleic acids to generate enriched candidate mixture. The SELEX Patent Applications also describe ligands obtained to a number of target species, including both protein targets where the protein is and is not a nucleic acid binding protein.

The methods described herein and the nucleic acid ligands identified by such methods are useful for both therapeutic and diagnostic purposes. Therapeutic uses include the treatment or prevention of diseases or medical conditions in human patients. Therapeutic uses may also include veterinary applications.

Diagnostic utilization may include both *in vivo* or *in vitro* diagnostic applications. The SELEX method generally, and the specific adaptations of the SELEX method taught and claimed herein specifically, are particularly suited for diagnostic applications. SELEX identifies nucleic acid ligands that are able to bind targets with high affinity and with surprising specificity. These characteristics are, of course, the desired properties one skilled in the art would seek in a diagnostic ligand.

The nucleic acid ligands of the present invention may be routinely adapted for diagnostic purposes according to any number of techniques employed by those skilled in the art. Diagnostic agents need only be able to allow the user to identify the presence of a given target at a particular locale or concentration. Simply the ability to form binding pairs with the target may be sufficient to trigger a positive signal for diagnostic purposes. Those skilled in the art

would also be able to adapt any nucleic acid ligand by procedures known in the art to incorporate a labeling tag in order to track the presence of such ligand. Such a tag could be used in a number of diagnostic procedures. The nucleic acid ligands to IgE described herein may specifically be used for identification of the IgE protein.

SELEX provides high affinity ligands of a target molecule. This represents a singular achievement that is unprecedented in the field of nucleic acids research. The present invention applies the SELEX procedure to the specific target of IgE. In the Example section below, the experimental parameters used to isolate and identify the nucleic acid ligands to IgE are described.

In order to produce nucleic acids desirable for use as a pharmaceutical, it is preferred that the nucleic acid ligand (1) binds to the target in a manner capable of achieving the desired effect on the target; (2) be as small as possible to obtain the desired effect; (3) be as stable as possible; and (4) be a specific ligand to the chosen target. In most situations, it is preferred that the nucleic acid ligand have the highest possible affinity to the target.

In co-pending and commonly assigned U.S. Patent Application Serial No. 07/964,624, filed October 21, 1992 ('624), methods are described for obtaining improved nucleic acid ligands after SELEX has been performed. The '624 application, entitled Methods of

Producing Nucleic Acid Ligands, is specifically incorporated herein by reference.

In the present invention, SELEX experiments were performed in order to identify RNA and ssDNA with specific high affinity for IgE from a degenerate library containing 40 or 60 random positions (40N or 60N). This invention includes the specific RNA ligands to IgE shown in Table 1 (SEQ ID NOS:7-41 and 43-48), identified by the methods described in Examples 1-3. This invention also includes the specific ssDNA ligands to IgE shown in Tables 5 and 6 (SEQ ID NOS:51-111), identified by the methods described in Examples 6, 7 and 10. The scope of the ligands covered by this invention extends to all nucleic acid ligands of IgE, modified and unmodified, identified according to the SELEX procedure. More specifically, this invention includes nucleic acid sequences that are substantially homologous to the ligands shown in Tables 1, 5 and 6. By substantially homologous it is meant a degree of primary sequence homology in excess of 70%, most preferably in excess of 80%. A review of the sequence homologies of the ligands of IgE shown in Tables 1, 5 and 6 show that sequences with little or no primary homology may have substantially the same ability to bind IgE. For these reasons, this invention also includes nucleic acid ligands that have substantially the same ability to bind IgE as the nucleic acid ligands shown in Tables 1, 5 and 6. Substantially the same ability to bind IgE means that the affinity is within a few orders of magnitude of the affinity of the ligands described herein. It is well within the skill

of those of ordinary skill in the art to determine whether a given sequence -- substantially homologous to those specifically described herein -- has substantially the same ability to bind IgE.

This invention also includes the ligands as described above, wherein certain chemical modifications are made in order to increase the *in vivo* stability of the ligand or to enhance or mediate the delivery of the ligand. Examples of such modifications include chemical substitutions at the sugar and/or phosphate and/or base positions of a given nucleic acid sequence. See, *e.g.*, U.S. Patent Application Serial No. 08/117,991, filed September 9, 1993, entitled High Affinity Nucleic Acid Ligands Containing Modified Nucleotides which is specifically incorporated herein by reference. Other modifications are known to one of ordinary skill in the art. Such modifications may be made post-SELEX (modification of previously identified unmodified ligands) or by incorporation into the SELEX process.

The nucleic acid ligands to the IgE protein described herein are useful as pharmaceuticals and as diagnostic reagents.

The following Examples are provided to explain and illustrate the present invention and are not intended to be limiting of the invention.

Example 1 describes the various experimental procedures used in Examples 2-5. Example 2 describes a representative method for identifying RNA ligands by the SELEX method which bind IgE and determines the affinities the ligands have for IgE. Example 3 maps

which regions of the ligands are necessary for IgE binding. Example 4 demonstrates the specificity of the ligands for Human IgE. Example 5 demonstrates that the ligands of the invention are capable of inhibiting the interaction between IgE and the FcεRI receptor. Example 6 describes the various experimental procedures used in Examples 7-10. Example 7 describes a representative method for identifying DNA ligands by the SELEX method which bind human IgE and presents the sequences and affinities the ligands have for IgE. Example 8 demonstrates the specificity of the ligands for Human IgE. Example 9 demonstrates that the DNA ligands are capable of inhibiting the interaction between IgE and the FcεRI receptor. Example 10 describes a representative method for identifying DNA ligands by the SELEX method which bind mouse IgE, presents the sequences, affinities, and specificity of the DNA ligands.

EXAMPLE 1. EXPERIMENTAL PROCEDURES

The experimental procedures provided in this example will be used in subsequent examples and are provided here to streamline the description.

A. Materials.

Human IgE used in this SELEX procedure was purchased from Athens Research Technology (Athens, GA); Mouse IgE was purchased from PharMingen (San Diego, CA). Biotinylated TAN IgE and the RBL cell lines, untransfected or transfected with the FcεRI receptor were a generous gift from Dr. Kinet (NIH) (Bethesda,

MD). 2' NH_2 modified CTP and UTP were prepared according to the method of Pieken *et al.* (1991) Science 253:314-317. DNA oligonucleotides were synthesized by Operon Technologies (Alameda, CA). All other reagents and chemicals were purchased from standard commercial sources.

B. SELEX.

The SELEX procedure has been described in detail in U.S. Patent No. 5,270,163 (see also Tuerk and Gold (1990) Science 24:505-510). For the IgE experiments, the DNA templates were designed to contain 40 or 60 random nucleotides, flanked by 5' and 3' regions of fixed structure (shown in Table 2) designated 40N7 (SEQ ID NO:1) and 60N7 (SEQ ID NO:2), respectively. The fixed regions include DNA primer annealing sites for PCR and cDNA synthesis as well as the consensus T7 promoter region to allow *in vitro* transcription. Single-stranded DNA molecules were converted into double-stranded transcribable templates by PCR amplification. PCR conditions were 50 mM KCl, 10 mM Tris-Cl, pH 9, 0.1% Triton X-100, 3 mM $MgCl_2$, 0.5 mM of each dATP, dCTP, dGTP, and dTTP, and contained 0.1 units/μl of Taq DNA polymerase. Transcription reactions contained 5μM DNA template, 5 units/μl T7 RNA polymerase, 40 mM Tris-Cl (pH 8.0), 12 mM $MgCl_2$, 5 mM DTT, 1 mM spermidine, 0.002% Triton X-100, 4% PEG 8000, 2 mM each of 2'-OH ATP, 2'-OH GTP, 2'-NH_2 CTP, 2'-NH_2 UTP, and 0.25 μM α-^{32}P 2'OH ATP. The RNA molecules were incubated with IgE protein in modified phosphate buffered saline (PBS), modified to contain 1 mM Mg^{2+}

ions, (138 mM NaCl, 2.7 mM KCl, 8.1 mM Na_2HPO4, 1.1 mM KH_2PO4, 1 mM $MgCl_2$, pH 7.4) for 10 min to allow binding to occur. IgE-RNA complexes were separated from unbound RNA by nitrocellulose filter partitioning. Bound RNA was isolated from filters by phenol/urea extraction. The RNA was reverse transcribed into cDNA by AMV reverse transcriptase (AMV RT) at 48°C for 60 min in 50 mM Tris-Cl (pH 8.3), 60 mM NaCl, 6 mM $Mg(OAc)_2$, 10 mM DTT, 50 pmol DNA primer, 0.4 mM each of dNTPs, and 1 unit/μl AMV RT. PCR amplification of this cDNA resulted in approximately 500 pmol double-stranded DNA which was used to initiate the next round of SELEX.

C. Nitrocellulose Filter Partitioning.

For isolation of RNA molecules that bind tightly to IgE, the nitrocellulose filter partitioning method was used as described in the SELEX Patent Applications. Filter discs (nitrocellulose/cellulose acetate mixed matrix, 0.45 μm pore size, Millipore Corporation, Bedford, MA) were placed into a vacuum manifold and wetted with 5 ml of modified PBS buffer. ^{32}P labeled RNA pools were incubated with serial dilutions of IgE in modified PBS for 10 min at 37°C and aspirated through the filter discs which was followed immediately by a 5 ml modified PBS wash. The filter discs were air-dried and counted in a liquid scintillation counter (Beckmann Instruments, Palo Alto, CA).

To obtain equilibrium dissociation constants of RNA ligands to IgE the binding reaction:

$R.P \rightarrow R+P$ R = RNA
P = Protein
K_D = dissociation constant

is converted to an equation for the fraction of RNA bound at equilibrium:

$$q = (f/2R_T)\circ(P_T+R_T+K_D-((P_T+R_T+K_D)^2-4P_TR_T)^{1/2})$$

q = fraction of RNA bound
P_T = total protein concentration
R_T = total RNA concentration
f = retention efficiency of RNA-protein complexes

The average retention efficiency for RNA-IgE complexes on nitrocellulose filters is 0.8. K_Ds were determined by least square fitting of the data points using the KALEIDAGRAPH graphic program (Synergy Software, Reading, PA).

D. Cloning and Sequencing.

During the last round of SELEX, PCR of cDNA was performed with the primers shown in Table 2 which contain recognition sites for the restriction endonucleases HindIII (5' primer 5P7H (SEQ ID NO:5)) and BamHI (3' primer 3P7B (SEQ ID NO:6)). Using these restriction sites the DNA sequences were inserted directionally into the pUC19 vector. These recombinant plasmids were transformed into *E. coli* SURE strain (Stratagene, La Jolla, CA). Plasmid DNA was prepared with the CLEARCUT miniprep kit (Stratagene, La Jolla, CA) and about 80 clones were sequenced with the SEQUENASE sequencing kit (United States Biochemical Corporation, Cleveland, OH).

E. Ligand Truncation.

Truncation experiments were carried out to determine the minimal sequence necessary for high affinity binding of the RNA ligands to IgE. For 3' boundary determination, RNA ligands were 5' end-labeled with γ- ^{32}P-ATP using T4 polynucleotide kinase. 5' boundaries were established with 3' end-labeled ligands using α-^{32}P-pCp and T4 RNA ligase. After partial alkaline hydrolysis, radiolabeled RNA ligands were incubated with IgE at concentrations ranging from 1 nM to 150 nM and protein-bound RNA was separated by nitrocellulose partitioning. RNA truncates were analyzed on a high-resolution denaturing polyacrylamide gel. A ladder of radioactively labeled ligands terminating with G-residues was generated by partial RNase T1 digestion and was used as markers.

F. IgE Receptor Binding Assay.

A cell-based enzyme linked immunosorbent assay (ELISA) was used to measure the ability of the high-affinity RNA ligands to inhibit binding of IgE to the FcεRI receptor. Rat basophilic leukemia (RBL) cell-line SX-38 which expresses the α, β and γ subunits of the human FcεRI IgE receptor was plated at a concentration of 10^5 cells per well in 96 well flat-bottom microtiter plates. After overnight growth, the cells were washed three times in modified PBS and incubated for one hour at 37°C in 30 μl of 1.5 μg/ml biotinylated TAN IgE. The cells were washed 3 more times and incubated in 30 μl of 1/100 dilution of horseradish peroxidase conjugated to avidin (Molecular Probes, Eugene, OR) for

one hour at 37°C. After three final washes, 30 μl of 3,3',5,5'-tetramethylbenzidine (TMB) substrate (Kirkegaard and Perry Laboratories, Gaithersburg, MD) were added as a chromogenic substrate and the plate was immediately transferred to the microtiter plate reader (Bio-Tek Instruments, Winooski, VT) for analysis. The absorbance at 630 nM was measured every 60 sec over a time span of 15 min. The rate of color development is an indication of the extent of IgE binding to the FcεRI receptor and was determined from the linear part of absorbance versus time graphs (data not shown). To test the effectiveness of RNA ligands to inhibit IgE binding to the receptor, the biotinylated TAN IgE was pre-incubated with RNA at concentrations ranging from 10^{-5} M to 3.3×10^{-9} M for 30 min at 37°C. As control reactions addition of RNA or IgE were omitted (maximum rate and background rate, respectively). The inhibition activities of the RNA ligands were determined using the following formula:

$$I_B = (R_R - B_R) / (M_R - B_R) \cdot 100$$

I_B = relative percentage IgE bound
R_R = reaction rate
B_R = background rate
M_R = maximum rate

Substituting transfected RBL SX-38 cells with the untransfected parental cell-line resulted in reaction rates similar to the background rate (data not shown). Values for the inhibition constant (K_i) were calculated based on the following estimates: 10^5 cells per well and 10^5 receptors per cell in 30 μl volume, resulting in a receptor concentration of 5×10^{-10} M. The IgE

concentration in this assay was 8 x 10^{-12} M and the dissociation constant for the IgE/receptor complex is 10^{-10} M.

EXAMPLE 2. RNA LIGANDS TO IGE

A. SELEX.

In order to generate stable ligands to human IgE two SELEX experiments were performed with 2'-NH_2 pyrimidine modified RNA molecules using the methods described in Example 1. As shown in Table 2, these RNA pools differ in the number of random bases present in the central portion of the molecules: 40 nucleotides in the 40N7 (SEQ ID NO:1) SELEX and 60 nucleotides in the 60N7 (SEQ ID NO:2) SELEX experiment. The starting pools of 3 x 10^{14} RNA molecules (500 pmol) bind IgE with approximate affinities of greater than 50 μM. After 9 rounds of SELEX, the affinities of the evolving pools had improved about two orders of magnitude and did not shift further in subsequent rounds. Round 9 RNA was bulk sequenced and found to be non-random. Therefore, cDNA from this round was PCR amplified with primers containing restriction sites and cloned into pUC19. About 35 clones from each SELEX experiment were sequenced as shown in Table 1. The sequences were inspected by eye and analyzed using computer programs which perform alignments.

B. RNA Sequences.

25 out of 35 (40N7 SELEX) and 30 out of 34 (60N7 SELEX) sequenced clones were found to be unique. As shown in Table 1, these unique clones can be divided

into four classes: high-affinity IgE ligand Groups A (SEQ ID NOS:7-18) and B (SEQ ID NOS:19-28), a group of unrelated high-affinity IgE ligands called orphan sequences (SEQ ID NOS:29-41) and nitrocellulose binding ligand Group C (not shown). It is interesting to note that all groups contain members of both 40N7 (SEQ ID NO:1) and 60N7 (SEQ ID NO:2) sequences. This indicates that independent SELEX experiments can result in the isolation of essentially identical ligands.

To crudely screen the ligands for their ability to bind to IgE, 2-point K_Ds were determined for all clones (data not shown). Group C ligands containing the consensus motif: 5' G-G-G-(N_{3-5})-G-U/C-G-G-A/U-G-G-G-G 3' (SEQ ID NO:42) were found to have high affinities for nitrocellulose instead of IgE and were not further analyzed.

Group A and group B ligands show characteristic conserved consensus domains of 10 and 20 nucleotides, respectively, immediately adjacent to the 5' fixed region as shown in Table 1. The conserved domains are:

5' GUG UGA AUG GUG UUG UGA GG 3' (SEQ ID NO:43)

5' GUG UGG GGC G 3' (SEQ ID NO:44)

The remaining bases of the variable regions 3' to these conserved stretches show no observable sequence conservation and are, therefore, unlikely to contribute to the specific binding of IgE.

C. Affinities.

The RNA ligands of group A, group B and most orphan sequences are high affinity IgE ligands and show very little binding to nitrocellulose. Dissociation

constants for representative members of Group A and Group B as well as orphan sequences were determined by nitrocellulose filter binding experiments and the dissociation constants are listed in Table 3. Group A ligands show an average K_D of approximately 150 nM, Group B ligands had an average K_D of approximately 35 nM and orphan ligands had K_Ds ranging from 50 nM to 250 nM.

EXAMPLE 3. LIGAND TRUNCATION

To determine the minimal sequence information necessary for high-affinity binding to IgE, truncation analysis was performed as described in Example 1 on representative members of Group A (IGEL1.1 (SEQ ID NO:7) and IGEL31.1 (SEQ ID NO:18) and Group B (IGEL2.1 (SEQ ID NO:20) and IGEL48.1 (SEQ ID NO:27). Table 1 shows that the 3' boundaries of the truncated ligands (SEQ ID NOS:45 and 46) are located, as expected, precisely at the end of the conserved consensus sequence motifs (SEQ ID NO:43) and (SEQ ID NO:44), in the variable region. This concurs with the hypothesis that these conserved sequence motifs are necessary for tight binding to IgE, whereas the degenerate remainder of the variable regions and the 3' fixed regions are dispensable. The 5' boundary determinations revealed that the entire 5' fixed regions, with the possible exception of the first three guanidine residues, are involved in binding to IgE. For technical purposes, further experiments were conducted with truncated ligands consisting of the intact 5' fixed region and the consensus domain, called IGEL1.2 (SEQ ID NO:47)

(Group A truncate of IGEL1.1) and IGEL2.2 (SEQ ID NO:48) (Group B truncate of IGEL2.1). Since these truncates are not missing any 5' sequence information critical for transcription initiation, they transcribe efficiently and yield several hundred RNA molecules per DNA template in overnight *in vitro* transcription reactions (data not shown). Representative binding data for these truncated RNA ligands in direct comparison with their full-length counterparts are shown in Table 3. While the K_D of IGEL2.2 is essentially unaffected by the truncation, IGEL1.2 actually binds several fold better as a truncate compared to the full-length IGEL1.1 ligand. This result demonstrates that the removal of nucleotides which are not contributing to the binding of a protein can increase the affinity of this interaction, possibly by allowing a tighter fit of the RNA to the surface of the protein.

EXAMPLE 4. SPECIFICITY OF RNA LIGANDS TO HUMAN IGE

To test the specificity of the high-affinity interaction between the RNA ligands and human IgE, binding experiments were performed with different immunoglobulins as described in Example 1. Table 4 summarizes the results of these studies with rat, murine and a human-murine hybrid IgE. The K_Ds for murine and rat IgE are estimated to be greater than 5 μM which is at least two orders of magnitude higher than the corresponding dissociation constants to human IgE. Furthermore, IgE ligands do not bind significantly to human IgG, a different immunoglobulin isotype (data not shown). These results indicate that

the RNA ligands are very specific for human IgE and do not interact efficiently with related molecules.

In order to localize which part of the IgE molecule the RNA ligands are contacting, binding experiments were carried out with a hybrid IgE molecule. The light chains and the variable region of this TAN IgE are murine derived, whereas the constant portion of the heavy chains are human. As shown in Table 4, the affinities of ligands IGEL1.1 (SEQ ID NO:7) and IGEL1.2 (SEQ ID NO:47) are equivalent to those determined for the entirely human IgE. These results indicate that ligands IGEL1.1 and IGEL1.2 are contacting IgE at the F_c portion. This part of the IgE molecule also harbors the contact region with the FcεRI receptor, making it potentially possible to competitively inhibit IgE binding to the receptor.

EXAMPLE 5. INHIBITION OF THE IGE - FCεRI INTERACTION

To directly test the ability of the RNA ligands to competitively inhibit the binding of IgE to the FcεRI receptor, a tissue culture cell based assay was performed as described in Example 1. Briefly, rat basophilic leukemia (RBL) cells that are expressing the human FcεRI receptor on the cell surface were incubated with biotinylated IgE. Using streptavidin conjugated horseradish peroxidase, the relative amount of IgE bound to the receptor can be determined by measuring the rate of conversion of a chromogenic substrate. The rate in the absence of inhibiting RNA is defined as 100% activity and the rate in the absence of IgE is used to calculate the background which is subtracted

from the conversion rates. The RNA species to be tested for competitive inhibition were pre-incubated with the IgE before the mixture was added to the RBL cells. Random RNA, the 40N7 (SEQ ID NO:1) and 60N7 (SEQ ID NO:2) starting pools and IGEL NC1, a scrambled version of ligand IGEL 1.2, do not show any significant inhibition of IgE binding to the FceRI receptor (data not shown). However, Group A ligands IGEL1.1 (SEQ ID NO:7) and IGEL1.2 (SEQ ID NO:47) do inhibit this interaction as shown in Table 4. The inhibitory dissociation constants (K_i) determined in this assay, 44 nM for IGEL1.1 and 21 nM for IGEL1.2, correspond well with the previously calculated K_D values of 77 nM and 36 nM, respectively, of these ligands for TAN IgE. These results show that binding of Group A ligands to IgE causes competitive inhibition of the interaction with the FceRI receptor. The orphan ligands tested in this assay show varying degrees of inhibition and most of them appear to be weaker inhibitors than IGEL1.1 and IGEL1.2. Group B ligands IGEL2.1 (SEQ ID NO:20) and IGEL2.2 (SEQ ID NO:48) do not show significant inhibition (data not shown) although they bind IgE tightly. This observation indicates that Group A and Group B ligands either bind IgE at different areas or that the binding orients the Group B ligands differently as to not allow inhibition of the IgE/receptor interaction.

EXAMPLE 6: EXPERIMENTAL PROCEDURES

A. Materials.

Materials used for Examples 6 - 10 are the same as those described in Example 1A.

B. SELEX.

The SELEX procedure has been described in detail in U.S. patent 5,270,163 (see also Tuerk and Gold (1990) Science 249; 505-510). The DNA template for the DNA SELEX experiments, 40NBHI, (Table 2; SEQ ID NO:112) was designed to contain 40 random nucleotides and is flanked by 5' and 3' regions of defined sequence used as primer annealing sites for PCR amplification (Table 2; SEQ ID NOS:49-50). Conditions for PCR amplifications were 2.5 μM 5' primer, 2.5 μM 3' primer, 50 mM KCl, 10 mM Tris-Cl, pH 9, 0.1% Triton X-100, 3 mM $MgCl_2$, 0.5 mM of each dATP, dCTP, dGTP, and dTTP, and contained 0.1 U/μl of Taq DNA polymerase.

Since the 3' primer contains two biotin molecules, the two DNA strands differ in molecular weight after amplification and were separated on denaturing polyacrylamide gels. To radioactively label the DNA molecules, 100 - 500 pmol of purified single stranded DNA were incubated in 50 mM Tris (pH 7.6), 10 mM $MgCl_2$, 5 mM dithiothreitol, 0.1 mM spermidine, 0.1 mM EDTA, 1.25 μM γ-^{32}P ATP with 0.2 U/μl T4 polynucleotide kinase at 37°C for 1 hour.

The 5' labeled DNA molecules were incubated with IgE in modified phosphate buffered saline (PBS), modified to contain 1 mM Mg^{2+} ions, (138 mM NaCl, 2.7 mM KCl, 8.1 mM NA_2HPO4, 1.1 mM KH_2PO4, 1 mM $MgCl_2$, pH

7.4) for 10 min to allow binding to occur. Protein-DNA complexes were separated from unbound DNA by nitrocellulose filter partitioning and bound DNA was isolated from filters by phenol/urea extraction.

C. Nitrocellulose Filter Partitioning.

For isolation of DNA molecules that bind tightly to IgE the nitrocellulose filter partitioning method was used as described in the SELEX Patent Applications. Filter discs (nitrocellulose/cellulose acetate mixed matrix, 0.45 μm pore size, Millipore Corporation, Bedford, MA) were placed into a vacuum manifold and pre-washed by aspiration of 5 ml of modified PBS buffer. ^{32}P labeled DNA pools were incubated with serial dilutions of IgE in modified PBS for 10 min at 37°C and aspirated through the filter discs which was followed immediately by a 5 ml modified PBS wash. The filter discs were air-dried and counted in a liquid scintillation counter (Beckmann Instruments, Palo Alto, CA).

To obtain equilibrium dissociation constants of DNA ligands to IgE the binding reaction:

$$D \cdot P \xrightarrow{K_D} D + P$$

D= DNA

P = Protein

K_D = dissociation constant

is converted to an equation for the fraction of DNA bound at equilibrium:

$$q = (f / 2D_T) \cdot (P_T + D_T + K_D - ((P_T + D_T + K_D)^2 - 4\ P_T\ D_T)^{1/2})$$

q = fraction of DNA bound

P_T = total protein concentration

D_T = total DNA concentration

f = retention efficiency of DNA-protein complexes

The average retention efficiency for DNA-IgE complexes on nitrocellulose filters is 0.5. K_Ds were determined by least square fitting of the data points using the graphic program Kaleidagraph (Synergy Software, Reading, PA).

D. Cloning and Sequencing.

During the last round of SELEX PCR amplification of the DNA pools was performed with primers which contain recognition sites for the restriction endonucleases HindIII (5' primer) (SEQ ID NO:5) and BamHI (3' primer) (SEQ ID NO: 6). Using these restriction sites, the DNA sequences were inserted directionally into the pUC9 vector. These recombinant plasmids were transformed into E. coli strain DH5a.

Plasmid DNA was prepared with the PERFECT prep kit (5' -> 3', Boulder, CO) and about 40 clones of each DNA SELEX experiment were sequenced with the Sequenase sequencing kit (United States Biochemical Corporation, Cleveland, OH).

F. IgE Receptor Binding Assay.

A cell-based enzyme linked immunosorbent assay (ELISA) was used to measure the ability of the high-affinity DNA ligands to inhibit binding of IgE to the FcεRI receptor. Rat basophilic leukemia (RBL) cell-line SX-38 which expresses the α, β and γ subunits of the

human FcεRI IgE receptor was plated at a concentration of 10^5 cells per well in 96 well flat-bottom microtiter plates. After overnight growth, the cells were washed three times in modified PBS and incubated for one hour at 37°C in 30 μl of 1.5 μg/ml biotinylated TAN IgE. The cells were washed 3 more times and incubated in 30 μl of 1/100 dilution of horseradish peroxidase (Molecular Probes, Eugene,OR) conjugated to avidin for one hour at 37°C. After three final washes, 30 μl of 3,3',5,5'-tetramethylbenzidine (TMB) substrate, (Kirkegaard and Perry Laboratories, Gaithersburg, MD) were added as a chromogenic substrate, and the plate was immediately transferred to the microtiter plate reader (Bio-Tek Instruments, Winooski, VT) for analysis. The absorbance at 630 nM was measured every 60 seconds over a time span of 15 minutes. The rate of color development is an indication of the extent of IgE binding to the FcεRI receptor and was determined from the linear part of absorbance versus time graphs (data not shown). To test the effectiveness of DNA ligands to inhibit IgE binding to the receptor, the biotinylated TAN IgE was pre-incubated with DNA at concentrations ranging from 10^{-5} M to 3.3×10^{-9} M for 30 min at 37°C. As control reactions, addition of DNA or IgE were omitted (maximum rate and background rate, respectively). The inhibition activities of the RNA

ligands were determined using the following formula:

$$I_B = (R_R - B_R) / (M_R - B_R) \times 100$$

I_B = relative percentage IgE bound

R_R = reaction rate

B_R = background rate

M_R = maximum rate

Substituting transfected RBL SX-38 cells with the untransfected parental cell-line resulted in reaction rates similar to the background rate (data not shown). Values for the inhibition constant (K_i) were calculated based on the following estimates: 10^5 cells per well and 10^5 receptors per cell in 30 μl volume, resulting in a receptor concentration of 5×10^{-10} M. The IgE concentration in this assay was 8×10^{-12} M and the dissociation constant for the IgE / receptor complex is 10^{-10} M.

EXAMPLE 7: DNA LIGANDS TO HUMAN IGE

A. SELEX.

In order to generate ligands to human IgE, a SELEX experiment was performed with DNA molecules using the methods described in Example 6. 2 nmole (1.2×10^{-15} molecules) of 40NBHI single stranded DNA were used to start SELEX against human IgE. The concentration of human IgE was 10^{-6} M in the first round and was gradually reduced during subsequent rounds to 5×10^{-10} M in round 15. The DNA concentration was kept at a 10 fold excess in the first round and at a 2 fold excess in subsequent rounds. These conditions resulted in a retention of 0.1 to 6% of the total DNA on

nitrocellulose filters. Binding of DNA to the filters in the absence of IgE was 10 to 400 fold lower. DNA molecules from round 15 were cloned into pUC9 and sequence information was obtained for 87 clones as shown in Table 5. The sequences were inspected by eye and aligned to reveal homologies.

B. Sequences.

52 out of 87 sequenced clones were found to be unique. As shown in Table 5, these clones can be grouped into one family characterized by a conserved sequence motif which is 21 bases in length and is located in the 40 nucleotide random region. This motif can be folded into a stem - loop structure. Bases flanking the consensus sequence can basepair with each other, therefore extending the conserved stem by an additional 2 to 11 basepairs. Extensive covariation in this DNA ligand family indicates that no specific primary sequence is required in this stem extension. To determine the minimal sequence requirements for high affinity binding to human IgE, three single stranded DNA species were synthesized: D17.0 (SEQ ID NO:57), the full-length ligand of clone 17 (80 nucleotides); D17.1 (SEQ ID NO:93) containing the 21 base conserved motif with two times four flanking bases forming the stem extension (29 nucleotides); and 17.4 (SEQ ID NO:94) which extends D17.1 with four 5' G residues and four 3' C residues (37 nucleotides).

C. Affinities.

Affinities of representative DNA ligands for human IgE were determined by nitrocellulose filter binding experiments and the K_Ds are listed in Table 7. During the 15 rounds of SELEX, the affinity of the DNA pool shifted more than three orders of magnitude from greater than 50 μM to 10 nM. Individual clones tested from this pool have affinities in the same range. The affinities were not expected to vary widely since the enriched DNA pool is very homogenous. Ligand D17.0 was picked for truncation experiments. Compared to the full-length ligand, the 29 base truncate D17.1, containing the conserved sequence motif and the 4 basepair extended stem, binds with a 8 fold weaker affinity to IgE. However, the affinity could be restored to the level of the full-length ligand by the addition of a 4 basepair stem resulting in the 37 base truncate D17.4. It is possible that the stem in D17.1 is not stable enough to prevent the formation of an alternative secondary structure and that the additional basepairs in D17.4 lock in the conformation necessary for IgE binding.

EXAMPLE 8: SPECIFICITY OF DNA LIGANDS TO HUMAN IGE

To test the specificity of the high-affinity interaction between the DNA ligands and human IgE, binding experiments were performed with different immunoglobulins. K_Ds of ligand D17.4 for mouse IgE and human IgG were determined to be greater than 10 μM which is at least three orders of magnitude higher than the corresponding dissociation constants to human IgE.

These result indicate that this DNA ligand is very specific for human IgE and does not bind efficiently to related molecules.

EXAMPLE 9: INHIBITION OF THE IGE - FcεRI INTERACTION

To test the ability of the DNA ligands to competitively inhibit the binding of IgE to the FcεRI receptor, a tissue culture cell based assay was performed. Rat basophilic leukemia (RBL) cells that are expressing the human FcεRI receptor on the cell surface were incubated with biotinylated IgE. Using streptavidin conjugated horseradish peroxidase, the relative amount of IgE bound to the receptor was determined by measuring the rate of conversion of a chromogenic substrate. The rate in the absence of inhibiting DNA is defined as 100% activity and the rate in the absence of IgE is used to calculate the background which is subtracted from the conversion rates. The DNA species to be tested for competitive inhibition were pre-incubated with the IgE before the mixture was added to the RBL cells. Random DNA, the 40NBHI starting pool (SEQ ID NO:112) and D17NC, the randomized version of D17.4, do not show any significant inhibition of IgE binding to the FcεRI receptor. In contrast, DNA ligands D17.0 (SEQ ID NO:57) and D17.4 (SEQ ID NO:94) inhibit this interaction with estimated inhibitory dissociation constants (K_i) of approximately 40 nM. These values are in the same range as the previously calculated K_D values of 10nM indicating that binding of D17.0 and

D17.4 ligands to IgE causes competitive inhibition of the interaction with the FcεRI receptor.

EXAMPLE 10: DNA LIGANDS TO MOUSE IGE

A. SELEX.

In order to generate ligands to mouse IgE, a SELEX experiment was performed with DNA molecules using the methods described in Example 6. DNA ligands to mouse IgE are useful for developing mouse models of allergic disease. 2 nmole (1.2×10^{-15} molecules) of 40NBHI single stranded DNA were used to start SELEX against mouse IgE. The concentration of mouse IgE was 10^{-6} M in the first round and was graduately reduced during subsequent rounds to 5×10^{-10} M in round 15. The DNA concentration was kept at a 10 fold excess in the first round and at a 2 fold excess in subsequent rounds. These conditions resulted in a retention of 0.2 to 2.6% of the total DNA on nitrocellulose filters. Binding of DNA to the filters in the absence of IgE was 8 to 200 fold lower. DNA molecules from round 15 were cloned into pUC9 and sequence information was obtained for 24 clones as described in Example 6 A-D.

B. Sequences, Affinities, and Specificity of DNA ligands.

16 out of 24 sequenced clones were found to be unique (Table 6; SEQ ID NOS:95-111). Despite extensive inspection by eye and analysis with the help of alignment programs no obvious sequence families were identified. The ligands showing similarity are believed to be clonally derived PCR artifacts rather

than independently isolated sequences. Furthermore, no similarities to the human IgE DNA ligands were identified. The affinities of the starting DNA pool and round 15 DNA pool for mouse IgE were determined by nitrocellulose filter binding experiments. During the SELEX experiment, the K_D of the DNA pool shifted more than 50 fold from greater than 1 μM to 20 nM. The round 15 DNA pool enriched for mouse IgE ligands was tested for binding to human IgE and the K_D was found to be greater than 50 μM. Therefore, the mouse IgE ligands do not bind significantly better to human IgE than random DNA.

TABLE 1
IgE Binding Ligands

*	**	SEQUENCE***
		Group A Sequences
7	1.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUUGUGAGGUUACUGUACUUCGGUGGCUGCAGACGACUCGCCCGA
8	21.1	GGGAGGACGAUGCGGGUGUGAACGGUGUUGUGAGGUUACUGUACUUCGGUGGCUGCAGACGACUCGCCCGA
9	22.1	GGGAGGACGAUGCGGGUGCGAAUGGUGUUGUGAGGAGCCUAAAUACGCGAUUGGUCAGACGACUCGCCCGA
10	23.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUUGUGAGGACUCGGAAGUUCCCCAGGGCCAGACGACUCGCCCGA
11	24.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUUGCGAGGCAUGCAGGAGGCGCUGUGGUCAGACGACUCGCCCGA
12	25.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUCGUGAGGACUUAUCAGGCUCCGUGGUGCAGACGACUCGCCCGA
13	26.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUUGUGAGGUUACUGCACUUCGGCGCUCAGACGACUCGCCCGA
14	27.1	GGGAGGACGAUGCGGGUGUGCAUGGUGUUGUGAGGCUGAGUAUAGGGGCCUGCGUCAGACGACUCGCCCGA
15	28.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUCGUGAGGAUGGAUUCGACAUGAGCGAUCAGACGACUCGCCCGA
16	29.1	GGGAGGACGAUGCGGGUGUCAAUGGUGUUGCGAGGCAAAAAUAACCAGCGCAUAUUCUCGGCCAUGUUGGCGUGCAUACAGACGACUCGC CCGA
17	30.1	GGGAGGACGAUGCGGGUGCGAAUGGUGUUGUGAGGAGUGAAUAUAGGUGGAUACCCCUUAACAACUGCGUGGGUCAGACGACUCGCCCGA
18	31.1	GGGAGGACGAUGCGGGUGUGAAUGGUGUUGUGAGGUUCUCGACUGUUUGUGUCUAGCCGUACUUUAGCCUCGGCCAGACGACUCGCCCGA
43	Consensus	GUGUGAAUGGUGUUGUGAGG
45	Truncated	AGGACGAUGCGGGUGUGAAUGGUGUUGUGAGG
47	1.2	GGGAGGACGAUGCGGGUGUGAAUGGUGUUGUGAGG
		Group B Sequences
19	41.1	GGGAGGACGAUGCGGGUGAGGGGCGAAUGGAGAACAUGAGACAAGGAGAAUGCGGCAGACGACUCGCCCGA
20	2.1	GGGAGGACGAUGCGGGUGUGGGGCGAAUGAGAAACGUUACCAGGAAAUGCGACUGCAGACGACUCGCCCGA
21	42.1	GGGAGGACGAUGCGGGAGUGGGGCGAAGGUAAUGUUGAGACGAUGUAAGACUGGUCAGACGACUCGCCCGA
22	43.1	GGGAGGACGAUGCGGGCGUGGGGCGAUUCAUAUCAACUGCUUAAGGUCACGGGUCCAGACGACUCGCCCGA
23	44.1	GGGAGGACGAUGCGGGUGCGGGGCGAGUAUAUGAAACUUGGCUUGGUAAUGAUCAGAAGUAGUGAGAACUGGGUGCAGACGACUCGCCCGA
24	45.1	GGGAGGACGAUGCGGGAGUGGGGCGUAGGAUUUGCCACUUGGAUUUGGACAGUGAGCAUCAGAGUCAUCACCGCCAGACGACUCGCCCGA
25	46.1	GGGAGGACGAUGCGGGAGUGGGGCGGAAUAACUAUGUGUGCGUAAUUGUCCUGUCGCGGUGUCACGAACCUUGUGCAGACGACUCGCCCGA
26	47.1	GGGAGGACGAUGCGGGUGUGGGGCGGAUAAUGAGUGAACAGAGUGAAAUUCCAGCGUACGCAGACUGUGCUGUCGCAGACGACUCGCCCGA
27	48.1	GGGAGGACGAUGCGGGAGUGGGGCGAUGAGAGAGAUCAGAGAACUAGAAGUGAUACAAAAUCUGAGGUUGUUGCGCAGACGACUCGCCCGA
28	49.1	GGGAGGACGAUGCGGGUGUGGGGCGGAUAAUGAGUGAACAGAGUGAAAUUCCAGUGUAGCCAGACUGUGCUGUCGCAGACGACUCGCCCGA
44	Consensus	GUGUGGGGCG
46	Truncated	AGGACGAUGCGGGUGUGGGGCG
48	2.2	GGGAGGACGAUGCGGGUGUGGGGCG

TABLE 1 CONT.

		Orphan Sequences
29	3.1	GGGAGGACGAUGCGGUCCAUCGAUUAGGCGGUCGUGCUGGUGUAGUGUGUAGUGGCAGACGACUCGCCCGA
30	4.1	GGGAGGACGAUGCGGACGGUAGUCUGGUAGGCGCUGUGACGGCGAGAAUCCGGACCAGACGACUCGCCCGA
31	5.1	GGGAGGACGAUGCGGGUAUCGGUACGUGUUGGCUUGGGAAGGGGUCCGACGGUGCAGACGACUCGCCCGA
32	6.1	GGGAGGACGAUGCGGACGGUAGUCUGGUAGGCGCUGUGACGCCGAGAAUCCGGAUCAGACGACUCGCCCGA
33	7.1	GGGAGGACGAUGCGGAGACGCGUGAACACUAGUAUCACAGUUAAGGAUGCGCGCAGACGACUCGCCCGA
34	8.1	GGGAGGACGAUGCGGACCGCCCGUCGAGGGCUAGGCGUAGAGUCUAACCGGUGCCAGACGACUCGCCCGA
35	9.1	GGGAGGACGAUGCGGGCGGGGAACAGACGGCUCAGAGCGGCACGAUUGUCAGCCAGCAAUUAUAUCGUGUUGAUGCAGACGACUCGCCCGA
36	10.1	GGGAGGACGAUGCGGGCCACGCGUGGUUAGGGAUCGCGGACAGCACAAAUCGAAUUUGAUUCGCACCUGGACGGUCAGACGACUCGCCCGA
37	11.1	GGGAGGACGAUGCGGAACAGCACGAGUGUACCUAAGACAGGCGAUGGCACUCGUGGUCGAAAUCAUAUAGUGAUGCAGACGACUCGCCCGA
38	13.1	GGGAGGACGAUGCGGGACGGGGCGGAAUCAUGCAUGUGAGCGAACAGAGAGAGAGCCGUGUAUCCAUUCGUGGUCAGACGACUCGCCCGA
39	15.1	GGGAGGACGAUGCGGAGGCGACGAGGUGGACAGGGGUAGGGAAGAUCGUCUGAAGUAUGCGUCCUUCCAGCCCGUCAGACGACUCGCCCGA
40	16.1	GGGAGGACGAUGCGGUGGAGCCUUUAGGGGGAAUAGUUGGCAGAAUUGCAGCUCAUGUAAUCUCGACUGUGUGUGCAGACGACUCGCCCGA
41	17.1	GGGAGGACGAUGCGGUGGAGCCUUUAGGGGGAUCGCACCUGAUCAAAGACGCAGUAUCGAUAGACUUGCGUGCCCAGACGACUCGCCCGA

* SEQ ID NO

** IGEL numbers

*** nucleotide abbreviations C and U actually depict the modified nucleotides 2'-NH_2-C and 2'-NH_2-U

Table 2

SEQ ID NO:	Nucleic Acid Sequence
	Starting RNAs For RNA SELEX Experiments:
	40N7:
1	5' GGGAGGACGAUGCGG [-40N-] CAGACGACUCGCCCGA 3'
	60N7:
2	5' GGGAGGACGAUGCGG [-60N-] CAGACGACUCGCCCGA 3'
	SELEX PCR Primers For RNA SELEX Experiments:
	5P7:
3	5' TAATACGACTCACTATAGGGAGGACGATGCGG 3'
	3P7:
4	5' TCGGGCGAGTCGTCTG 3'
	Cloning PCR Primers For RNA SELEX Experiments:
	5P7H:
5	5' CCGAAGCTTAATACGACTCACTATAGGGAGGACGATGCGG 3'
	3P7B:
6	5' GCCGGATCCTCGGGCGAGTCGTCTG 3'
49	5' primer for DNA SELEX experiments:
	5BH1:
	5' CTACCTACGATCTGACTAGC 3'
50	3' primer for DNA SELEX experiments:
	3BBH1:
	5' AJAJAGGAACTACATGAGAGTAAGC 3'
112	Starting DNA:
	40NBH1
	5' CTACCTACGATCTGACTAGC [-40N-] GCTTACTCTCATGTAGTTCC 3'

Table 3

Group	SEQ ID NO:	Ligand	Dissociation Constant
A	7	IGEL1.1	138 nM
	12	IGEL25.1	186 nM
	16	IGEL29.1	141 nM
	18	IGEL31.1	134 nM
B	20	IGEL2.1	41 nM
	23	IGEL44.1	34 nM
	27	IGEL48.1	30 nM
	28	IGEL49.1	39 nM
Orphans	29	IGEL3.1	56 nM
	30	IGEL4.1	87 nM
	31	IGEL5.1	145 nM
	33	IGEL7.1	142 nM
	34	IGEL8.1	225 nM
	35	IGEL9.1	105 nM
	36	IGEL10.1	69 nM
	37	IGEL11.1	129 nM
	38	IGEL13.1	50 nM
Group A Truncate	47	IGEL1.2	19 nM
Group B Truncate	48	IGEL2.2	44 nM

Table 4

Protein	**Ligand (SEQ ID NO:)**	**Dissociation Constant**	**Inhibition Constant**
Murine IgE	IGEL1.2 (47)	≥ 5 μM	
	IGEL2.2 (48)	≥ 5 μM	
Rat IgE	IGEL1.2 (47)	≥ 5 μM	
	IGEL2.2 (48)	≥ 5 μM	
TAN IgE	IGEL1.1 (47)	77 nM	44 nM
	IGEL1.2 (48)	36 nM	21 nM

Table 5

Human IgE

51	D151	ctacctacgatctgactagc TACCCGCGATGAGAGTAAGTTTATCCGTGTACTCTTAGTG gcttactctcatgtagttcc
52	D161	ctacctacgatctgactagc TACCCGCGTTGAGAGTAAGTTTATCCGTGTACTCTTAGTG gcttactctcatgtagttcc
53	D066	ctacctacgatctgactagc ACAGCATGAGAGATATAGCTTTATCCGTGACTCTCAGTGG gcttactctcatgtagttcc
54	D160	ctacctacgatctgactagc CAATTGCTGAAGGAAGCATTTATCCGTTCCTCTTAGTGGT gcttactctcatgtagttcc
55	D016	ctacctacgatctgactagc CAACTGCTGAAGGAAGCATTTATCCGTTCCTCTTAGTGGT gcttactctcatgtagttcc
56	D054	ctacctacgatctgactagc AGGTCATGCGAGTATGCTTTATCCGTAACCTCTCAGTGG gcttactctcatgtagttcc
57	D017	ctacctacgatctgactagc CAATGAGTGTACCACGTTTATCCGTCCCTCCTAGTGGCGT gcttactctcatgtagttcc
58	D001	ctacctacgatctgactagc CCCTGANGTGTNMAMKTTTGTWCCGTTYCTCCTAGTGGCGT gcttactctcatgtagttcc
59	D069	ctacctacgatctgactagc GGCCGTAAGCAACCTTTATCCGTAATCTCTCAGTGGGGTA gcttactctcatgtagttcc
60	D165	ctacctacgatctgactagc GTGMGCGGGATCTTTATYCGTTACTCTTAGTGGGTCTCG gcttactctcatgtagttcc
61	D051	ctacctacgatctgactagc AAGGCGACTACTTTATCCGTTTCTCTTAGTGGGTATCCG gcttactctcatgtagttcc
62	D173	ctacctacgatctgactagc AATGGTCCAGCTTTATCCGTCTCTTTCAGTGGCGTCATT gcttactctcatgtagttcc
63	D012	ctacctacgatctgactagc CTACHGCCCATTTATVCGTTCCTCCTAGTGGTGGGCTGCT gcttactctcatgtagttcc
64	D019	ctacctacgatctgactagc RGCCGGGACATTTATCCGTTACTACTCAGTGGGTGAACTGTC gcttactctcatgtagttcc
65	D029	ctacctacgatctgactagc WCCGGAGTACTTTATYCGTYCCTTCTAGTGGGTACCCGTA gcttactctcatgtagttcc
66	D002	ctacctacgatctgactagc GGCCGGAGCTTTATCCGTTACTCTCAGTGGGTGACTGTC gcttactctcatgtagttcc

Table 5 cont.

67	D067	ctacctacgatctgactagc	GGCCGGAGCTTTATTCCGTTACTCTCAGTGGGTGACTGTC	gcttactctcatgtagttcc
68	D023	ctacctacgatctgactagc	CAAAGTTAATTTATNCGTCCCTCCTAGTGGTTAACAGCG	gcttactctcatgtagttcc
69	D153	ctacctacgatctgactagc	CAAAGTTAATTTATCCGTCCCTCTCAGTGGTTAACAGCG	gcttactctcatgtagttcc
70	D170	ctacctacgatctgactagc	CAAGGTTAATTTATYCGTCCCTCCCAGTGGTTAACAGCG	gcttactctcatgtagttcc
71	D059	ctacctacgatctgactagc	GATGGGAGCTTTATCCGTTCACTCTCAGTGGGCTCCTCAG	gcttactctcatgtagttcc
72	D072	ctacctacgatctgactagc	GATGGGAGCTTTATCCGTTCACTTTCAGTGGGCTCCTCAT	gcttactctcatgtagttcc
73	D053	ctacctacgatctgactagc	CGAAGTTAATTTATSCGTCCCTCCTAGTGGYTTAACAGCG	gcttactctcatgtagttcc
74	D172	ctacctacgatctgactagc	CTTGCTCCATTTATCCGTTTCTCCCAGTGGTGGTTGCATG	gcttactctcatgtagttcc
75	D061	ctacctacgatctgactagc	CTACGCGCTTTATCCGTTTCTCCCAGTGGGCGGGCGTTC	gcttactctcatgtagttcc
76	D080	ctacctacgatctgactagc	GTVSSGCTTTATCCGTTNCTCCCAGTGGGCGGGCRTTC	gcttactctcatgtagttcc
77	D169	ctacctacgatctgactagc	GGCTTTATCCGTAACCTCTTAGTGGGCCGCNCGCTTCACA	gcttactctcatgtagttcc
78	D009	ctacctacgatctgactagc	ATGGGAGAACACTTAGCCTTCATCCGTTCCTCCTAGTGGG	gcttactctcatgtagttcc
79	D052	ctacctacgatctgactagc	CGCGCGTACGAGCACCTTCATCCGTCCCTCCTAGTGGGGT	gcttactctcatgtagttcc
80	D074	ctacctacgatctgactagc	GGCCGTAASCAAGCCTTYWTCCGTMACCTACTYAGTGGGGKR	gcttactctcatgtagttcc
81	D159	ctacctacgatctgactagc	NNNNNNNNCTCTTTCATCCGTACCTCCCAGTGGAGAACGC	gcttactctcatgtagttcc
82	D011	ctacctacgatctgactagc	ACCGGAGTACTTCATCCGTCCCTTCTAGTGGGTACCCGTA	gcttactctcatgtagttcc
83	D073	ctacctacgatctgactagc	TCCGGAGTACTTCATCCGTYCCTTCTAGTGGGTACCCGTA	gcttactctcatgtagttcc

Table 5 cont.

84	D152	ctacctacgatctgactagc AGGGATGTTCATCCGTTCCTCTCAGTGGCATCCCGTGGCT gcttactctcatgtagttcc
85	D158	ctacctacgatctgactagc NNNNGGTTCATCCGTTCCTTCTAGTGGCCACCTGGATGCA gcttactctcatgtagttcc
86	D162	ctacctacgatctgactagc TGGCATTCATCCGTCTCTCCTAGTGGTGCCTTGTCCCCCA gcttactctcatgtagttcc
87	D058	ctacctacgatctgactagc CCTTCATCCGTTACTCTTAGTGGGGGCTTGCGATTCGAGT gcttactctcatgtagttcc
88	D056	ctacctacgatctgactagc TGCTGGACAATTGATCCGTTACTCTTAGTGGTTGTGTGCT gcttactctcatgtagttcc
89	D021	ctacctacgatctgactagc ACGGGTGAGTTGATCCGTTACTCTTAGTGGTGAACCTTGT gcttactctcatgtagttcc
90	D031	ctacctacgatctgactagc ACGGGTGAGTTGATCCGTCACTCTTAGTGGTGAACCTTGT gcttactctcatgtagttcc
91	D068	ctacctacgatctgactagc ACAGGTGAGTTGATCCGTTACTCTTAGTGGTGAACCTTGT gcttactctcatgtagttcc
92	D079	ctacctacgatctgactagc ACTGGTGAKTTGATCCGTCACTCTTAGTGGTGAWCCTTGT gcttactctcatgtagttcc
93	D17.1	CACGTTTATCCGTCCCTCCTAGTGGCGTG
94	D17.4	GGGGCACGTTTATCCGTCCCTCCTAGTGGCGTGCCCC

Table 6

Mouse IgE

95	D114	ctacctacgatctgactagc	GGCGTTTTACTTACTGGTCTTAGACCGGAGAGACACAGTC	gcttactctcatgtagttcc
96	D134	ctacctacgatctgactagc	GGCGTTTTACGTGTACKGGTCATGTGAGRCSGGAGAGACACRGTK	gcttactctcatgtagttcc
97	D109	ctacctacgatctgactagc	GATGGGNGGGGACGTGCTGATTTCCCACTTCATATTTCGT	gcttactctcatgtagttcc
98	D119	ctacctacgatctgactagc	GAGGGTGTGGGACGTGCTGATTTCCCACTTCATATTTCGT	gcttactctcatgtagttcc
99	D110	ctacctacgatctgactagc	CTGGGKKTCCCGCGGAKCKGTTACCAGTTATGGGGCAAG	gcttactctcatgtagttcc
100	D129	ctacctacgatctgactagc	CTGGGGGTSCCGCGGAGCGTGTTACCAGTTATGGGGCAAT	gcttactctcatgtagttcc
101	D131	ctacctacgatctgactagc	CTGGGGGTCCCGCGTAGCGTGTTACCAGTTATGGGGCAAT	gcttactctcatgtagttcc
102	D128	ctacctacgatctgactagc	CCTACCGTCGKCGCGGTTAAGGAAACTACBGCCTTTTACB	gcttactctcatgtagttcc
103	D101	ctacctacgatctgactagc	ACCGCTAGTTTCGAGGTTGGACGTGTTTGCCGTGTCGATT	gcttactctcatgtagttcc
104	D101	ctacctacgatctgactagc	ACCGCTAGTTTCGAGGTTGGACGTGTTTGCCGTGTCGATT	gcttactctcatgtagttcc
105	D102	ctacctacgatctgactagc	GTTGGACGGTTACGTTTCCTCATGGCAACCCAGCTAGATC	gcttactctcatgtagttcc
106	D112	ctacctacgatctgactagc	TTTCCCTCGACGGGTGCCCACTGCGGCATGGGTTAAGA	gcttactctcatgtagttcc
107	D113	ctacctacgatctgactagc	TGGGGCAGCTTTGCGNGGGTCCTACGTTTTACDTTTGCC	gcttactctcatgtagttcc
108	D121	ctacctacgatctgactagc	NNCGTCCTTCCAGTGGTGGAGTACCACCCGTCCGGCACTT	gcttactctcatgtagttcc
109	D124	ctacctacgatctgactagc	CGCGGGTGATCGGATTRCCCTGCATCTCCRCCTGATTCTT	gcttactctcatgtagttcc
110	D127	ctacctacgatctgactagc	GATNGCGTTTCGATACTGCTTCTGCGGAGTCACACAGCTC	gcttactctcatgtagttcc
111	D135	ctacctacgatctgactagc	AGGCGGGTNCCCTCTGGCGGAACACTTTGCTGTTGTCCT	gcttactctcatgtagttcc

Table 7

Ligand	K_D
40NBHI	≥ 50 μM
Round 15 human IgE	10 nM
Ligand enriched pool D2.0	6 nM
D11.0	8 nM
D31.0	22 nM
D17.0	9 nM
D17.1	82 nM
D17.4	10 nM
D17NC	≥ 50 μM

SEQUENCE LISTING

(1) GENERAL INFORMATION:

(i) APPLICANT: WIEGAND, TORSTEN
GOLD, LARRY
TASSET, DIANE

(ii) TITLE OF THE INVENTION: HIGH-AFFINITY OLIGONUCLEOTIDE LIGANDS TO IMMUNOGLOBIN E

(iii) NUMBER OF SEQUENCES: 112

(iv) CORRESPONDENCE ADDRESS:
(A) ADDRESSEE: Swanson & Bratschun, L.L.C.
(B) STREET: 8400 East Prentice Avenue, Suite 200
(C) CITY: Englewood
(D) STATE: Colorado
(E) COUNTRY: USA
(F) ZIP: 80111

(v) COMPUTER READABLE FORM:
(A) MEDIUM TYPE: Diskette, 3.5 inch, 1.44 MG storage
(B) COMPUTER: IBM compatible
(C) OPERATING SYSTEM: MS-DOS
(D) SOFTWARE: WordPerfect 6.0

(vi) CURRENT APPLICATION DATA:
(A) APPLICATION NUMBER: PCT/US95/
(B) FILING DATE:
(C) CLASSIFICATION:

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 08/471,985
(B) FILING DATE: 06-JUNE-1995

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 08/317,403
(B) FILING DATE: 03-OCTOBER-1994

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 07/714,131
(B) FILING DATE: 10-JUNE-1991

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 07/536,428
(B) FILING DATE: 11-JUNE-1990

(viii) ATTORNEY/AGENT INFORMATION:

(A) NAME: Barry J. Swanson
(B) REGISTRATION NUMBER: 33,215
(C) REFERENCE/DOCKET NUMBER: NEX23C/PCT

(ix) TELECOMMUNICATION INFORMATION:
(A) TELEPHONE: (303) 793-3333
(B) TELEFAX: (303) 793-3433

(2) INFORMATION FOR SEQ ID NO:1:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:1:
GGGAGGACGA UGCGGNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN 50
NNNNNCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:2:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:
GGGAGGACGA UGCGGNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN 50
NNNNNNNNNN NNNNNNNNNN NNNNNCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:3:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 32
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:3:

TAATACGACT CACTATAGGG AGGACGATGC GG 32

(2) INFORMATION FOR SEQ ID NO:4:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 16
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:4:
TCGGGCGAGT CGTCTG 16

(2) INFORMATION FOR SEQ ID NO:5:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 40
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:
CCGAAGCTTA ATACGACTCA CTATAGGGAG GACGATGCGG 40

(2) INFORMATION FOR SEQ ID NO:6:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 25
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:6:
GCCGGATCCT CGGGCGAGTC GTCTG 25

(2) INFORMATION FOR SEQ ID NO:7:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:7:
GGGAGGACGA UGCGGGUGUG AAUGGUGUUG UGAGGUUACU GUACUUCGGU 50
GGCUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:8:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:8:
GGGAGGACGA UGCGGGUGUG AACGGUGUUG UGAGGUUACU GUACUUCGGU 50
GGCUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:9:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:9:
GGGAGGACGA UGCGGGUGCG AAUGGUGUUG UGAGGAGCCU AAAUACGCGA 50
UUGGUCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:10:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:10:
GGGAGGACGA UGCGGGUGUG AAUGGUGUUG UGAGGACUCG GAAGUUCCCC 50
AGGGCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:11:

(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:11:

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GGGAGGACGA UGCGGGUGUG AAUGGUGUUG CGAGGCAUGC AGGAGGCGCU     50
GUGGUCAGAC GACUCGCCCG A                                    71
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(2) INFORMATION FOR SEQ ID NO:12:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:12:

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GGGAGGACGA UGCGGGUGUG AAUGGUGUCG UGAGGACUUA UCAGGCUCCG     50
UGGUGCAGAC GACUCGCCCG A                                    71
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(2) INFORMATION FOR SEQ ID NO:13:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:13:

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GGGAGGACGA UGCGGGUGUG AAUGGUGUUG UGAGGUUACU GCACUUCGGC     50
GCUCAGACGA CUCGCCCGA                                       71
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(2) INFORMATION FOR SEQ ID NO:14:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:14:

GGGAGGACGA UGCGGGUGUG CAUGGUGUUG UGAGGCUGAG UAUAGGGGCC 50
UGCGUCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:15:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:15:

GGGAGGACGA UGCGGGUGUG AAUGGUGUCG UGAGGAUGGA UUCGACAUGA 50
GCGAUCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:16:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 94
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:16:

GGGAGGACGA UGCGGGUGUC AAUGGUGUUG CGAGGCAAAA AUAACCAGCG 50
CAUAUUCUCG GCCAUGUUGG CGUGCAUACA GACGACUCGC CCGA 94

(2) INFORMATION FOR SEQ ID NO:17:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 90

(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:17:

GGGAGGACGA UGCGGGUGCG AAUGGUGUUG UGAGGAGUGA AUAUAGGUGG 50
AUACCCCUUA ACAACUGCGU GGGUCAGACG ACUCGCCCGA 90

(2) INFORMATION FOR SEQ ID NO:18:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 90
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:18:

GGGAGGACGA UGCGGGUGUG AAUGGUGUUG UGAGGUUCUC GACUGUUUGU 50
GUCUAGCCGU ACUUUAGCCU CGGCCAGACG ACUCGCCCGA 90

(2) INFORMATION FOR SEQ ID NO:19:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:19:

GGGAGGACGA UGCGGGUGAG GGCGAAUGG AGAACAUGAG ACAAGGAGAA 50
UGCGGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:20:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:20:
GGGAGGACGA UGCGGGUGUG GGGCGAAUGA GAAACGUUAC CAGGAAAUGC 50
GACUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:21:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:21:
GGGAGGACGA UGCGGGAGUG GGGCGAAGGU AAUGUUGAGA CGAUGUAAGA 50
CUGGUCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:22:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:22:
GGGAGGACGA UGCGGGCGUG GGGCGAUUCA UAUCAACUGC UUAAGGUCAC 50
GGGUCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:23:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single

(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:23:

GGGAGGACGA UGCGGGUGCG GGGCGAGUAU AUGAAACUUG GCUUGGUAAU 50
GAUCAGAAGU AGUGAGAACU GGGUGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:24:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 90
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:24:

GGGAGGACGA UGCGGGAGUG GGGCGUAGGA UUUGCCACUU GGAUUUGGAC 50
AGUGAGCAUC AGAGUCAUCA CCGCCAGACG ACUCGCCCGA 90

(2) INFORMATION FOR SEQ ID NO:25:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:25:

GGGAGGACGA UGCGGGAGUG GGGCGGAAUA ACUAUGUGUG CGUAAUUGUC 50
CUGUCGCGGU GUCACGAACC UUGUGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:26:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:26:

GGGAGGACGA UGCGGGUGUG GGGCGGAUAA UGAGUGAACA GAGUGAAAUU 50
CCAGCGUACG CAGACUGUGC UGUCGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:27:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:27:

GGGAGGACGA UGCGGGAGUG GGGCGAUGAG AGAGAUCAGA GAACUAGAAG 50
UGAUACAAAA UCUGAGGUUG UUGCGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:28:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:28:

GGGAGGACGA UGCGGGUGUG GGGCGGAUAA UGAGUGAACA GAGUGAAAUU 50
CCAGUGUAGC CAGACUGUGC UGUCGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:29:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:29:

GGGAGGACGA UGCGGUCCAU CGAUUAGGCG GUCGUGCUGG UGUAGUGUGU 50
AGUGGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:30:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:30:

GGGAGGACGA UGCGGACGGU AGUCUGGUAG GCGCUGUGAC GGCGAGAAUC 50
CGGACCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:31:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:31:

GGGAGGACGA UGCGGGUAUC GGUACGUGUU GGCUUGGGAA GGGGUCCGAC 50
GGUGCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQ ID NO:32:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:

(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:32:

GGGAGGACGA UGCGGACGGU AGUCUGGUAG GCGCUGUGAC GCCGAGAAUC 50
CGGAUCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQ ID NO:33:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:33:

GGGAGGACGA UGCGGAGACG CGUGAACACU AGUAUCACAG UUAAGGAUGC 50
GCGCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQ ID NO:34:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:34:

GGGAGGACGA UGCGGACCGC CCGUCGAGGG CUAGGCGUAG AGUCUAACCG 50
GUGCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQ ID NO:35:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine

(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:35:
GGGAGGACGA UGCGGGCGGG GAACAGACGG CUCAGAGCGG CACGAUUGUC 50
AGCCAGCAAU UAUAUCGUGU UGAUGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:36:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:36:
GGGAGGACGA UGCGGGCCAC GCGUGGUUAG GGAUCGCGGA CAGCACAAAU 50
CGAAUUUGAU UCGCACCUGG ACGGUCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:37:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:37:
GGGAGGACGA UGCGGAACAG CACGAGUGUA CCUAAGACAG GCGAUGGCAC 50
UCGUGGUCGA AAUCAUAUAG UGAUGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:38:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:

(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:38:
GGGAGGACGA UGCGGGACGG GGGCGGAAUC AUGCAUGUGA GCGAACAGAG 50
AGAGAGCCGU GUAUCCAUUC GUGGUCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:39:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:39:
GGGAGGACGA UGCGGAGGCG ACGAGGUGGA CAGGGGUAGG GAAGAUCGUC 50
UGAAGUAUGC GUCCUUCCAG CCCGUCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:40:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 91
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:40:
GGGAGGACGA UGCGGUGGAG CCUUUAGGGG GAAUAGUUGG CAGAAUUGCA 50
GCUCAUGUAA UCUCGACUGU GUGUGCAGAC GACUCGCCCG A 91

(2) INFORMATION FOR SEQ ID NO:41:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 90
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:41:

GGGAGGACGA UGCGGUGGAG CCUUUAGGGG GAUCGCACCU GAUCAAAGAC 50
GCAGUAUCGA UAGACUUGCG UGCCCAGACG ACUCGCCCGA 90

(2) INFORMATION FOR SEQ ID NO:42:

(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 13
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(ix) FEATURE:
(D) OTHER INFORMATION: N at position 4 is 3-5 nucleotides
(ix) FEATURE:
(D) OTHER INFORMATION: N at position 6 is U or C
(ix) FEATURE:
(D) OTHER INFORMATION: N at position 9 is A or U
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:42:

GGGNGNGGNG GGG 13

(2) INFORMATION FOR SEQ ID NO:43:

(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 20
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:43:

GUGUGAAUGG UGUUGUGAGG 20

(2) INFORMATION FOR SEQ ID NO:44:

(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 10
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:44:
GUGUGGGGCG 10

(2) INFORMATION FOR SEQ ID NO:45:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 32
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:45:
AGGACGAUGC GGGUGUGAAU GGUGUUGUGA GG 32

(2) INFORMATION FOR SEQ ID NO:46:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 22
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:46:
AGGACGAUGC GGGUGUGGGG CG 22

(2) INFORMATION FOR SEQ ID NO:47:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 35
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:47:
GGGAGGACGA UGCGGGUGUG AAUGGUGUUG UGAGG 35

(2) INFORMATION FOR SEQ ID NO:48:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 35
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All C's are 2'-NH_2 cytosine
(ix) FEATURE:
(D) OTHER INFORMATION: All U's are 2'-NH_2 uracil
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:48:
GGGAGGACGA UGCGGGUGUG GGGCG 35

(2) INFORMATION FOR SEQ ID NO:49:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 20 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:49:
CTACCTACGA TCTGACTAGC 20

(2) INFORMATION FOR SEQ ID NO:50:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 25 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(ix) FEATURE:
(D) OTHER INFORMATION: J at position 2 and 4 is Biotin
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:50:
AJAJAGGAAC TACATGAGAG TAAGC 25

(2) INFORMATION FOR SEQ ID NO:51:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:51:
CTACCTACGA TCTGACTAGC TACCCGCGAT GAGAGTAAGT TTATCCGTGT 50
ACTCTTAGTG GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:52:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:52:
CTACCTACGA TCTGACTAGC TACCCGCGTT GAGAGTAAGT TTATCCGTGT 50
ACTCTTAGTG GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:53:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:53:
CTACCTACGA TCTGACTAGC ACAGCATGAG AGATATAGCT TTATCCGTGA 50
CTCTCAGTGG GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:54:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:54:
CTACCTACGA TCTGACTAGC CAATTGCTGA AGGAAGCATT TATCCGTTCC 50
TCTTAGTGGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:55:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:55:
CTACCTACGA TCTGACTAGC CAACTGCTGA AGGAAGCATT TATCCGTTCC 50

TCTTAGTGGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:56:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:56:
CTACCTACGA TCTGACTAGC AGGTCATGCG AGTATGCTTT ATCCGTAACC 50
TCTCAGTGGG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:57:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:57:
CTACCTACGA TCTGACTAGC CAATGAGTGT ACCACGTTTA TCCGTCCCTC 50
CTAGTGGCGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:58:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 81 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:58:
CTACCTACGA TCTGACTAGC CCCTGANGTG TNMAMKTTTG TWCCGTTYCT 50
CCTAGTGGCG TGCTTACTCT CATGTAGTTC C 81

(2) INFORMATION FOR SEQ ID NO:59:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:59:
CTACCTACGA TCTGACTAGC GGCCGTAAGC AACCTTTATC CGTAATCTCT 50
CAGTGGGGTA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:60:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:60:

CTACCTACGA TCTGACTAGC GTGMGCGGGA TCTTTATYCG TTACTCTTAG 50
TGGGTCTCGG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:61:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:61:

CTACCTACGA TCTGACTAGC AAGGCGACTA CTTTATCCGT TTCTCTTAGT 50
GGGTATCCGG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:62:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:62:

CTACCTACGA TCTGACTAGC AATGGTCCAG CTTTATCCGT CTCTTTCAGT 50
GGGCGTCATT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:63:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:63:

CTACCTACGA TCTGACTAGC CTACHGCCCA TTTATVCGTT CCTCCTAGTG 50
GTGGGCTGCT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:64:
(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 82 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:64:

CTACCTACGA TCTGACTAGC RGCCGGGACA TTTATCCGTT ACTACTCAGT 50
GGGTGAACTG TCGCTTACTC TCATGTAGTT CC 82

(2) INFORMATION FOR SEQ ID NO:65:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:65:

CTACCTACGA TCTGACTAGC WCCGGAGTAC TTTATYCGTY CCTTCTAGTG 50
GGTACCCGTA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:66:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:66:

CTACCTACGA TCTGACTAGC GGCCGGAGCT TTATCCGTTA CTCTCAGTGG 50
GTGACTGTCG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:67:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:67:

CTACCTACGA TCTGACTAGC GGCCGGAGCT TTATTCCGTT ACTCTCAGTG 50
GGTGACTGTC GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:68:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:68:

```
CTACCTACGA TCTGACTAGC CAAAGTTAAT TTATNCGTCC CTCCTAGTGG    50
TTAACAGCGG CTTACTCTCA TGTAGTTCC                           79
```

(2) INFORMATION FOR SEQ ID NO:69:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:69:

```
CTACCTACGA TCTGACTAGC CAAAGTTAAT TTATCCGTCC CTCTCAGTGG    50
TTAACAGCGG CTTACTCTCA TGTAGTTCC                           79
```

(2) INFORMATION FOR SEQ ID NO:70:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:70:

```
CTACCTACGA TCTGACTAGC CAAGGTTAAT TTATYCGTCC CTCCCAGTGG    50
TTAACAGCGG CTTACTCTCA TGTAGTTCC                           79
```

(2) INFORMATION FOR SEQ ID NO:71:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:71:

```
CTACCTACGA TCTGACTAGC GATGGGAGCT TTATCCGTTC ACTCTCAGTG    50
GGCTCCTCAG GCTTACTCTC ATGTAGTTCC                          80
```

(2) INFORMATION FOR SEQ ID NO:72:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:72:
CTACCTACGA TCTGACTAGC GATGGGAGCT TTATCCGTTC ACTTTCAGTG 50
GGCTCCTCAT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:73:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:73:
CTACCTACGA TCTGACTAGC CGAAGTTAAT TTATSCGTCC CTCCTAGTGG 50
YTTAACAGCG GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:74:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:74:
CTACCTACGA TCTGACTAGC CTTGCTCCAT TTATCCGTTT CTCCCAGTGG 50
TGGTTGCATG GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:75:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:75:
CTACCTACGA TCTGACTAGC CTACGCGCTT TATCCGTTTC TCCCAGTGGG 50
CGGGCGTTCG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:76:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 78 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:76:

CTACCTACGA TCTGACTAGC GTVSSGCTTT ATCCGTTNCT CCCAGTGGGC 50
GGGCRTTCGC TTACTCTCAT GTAGTTCC 78

(2) INFORMATION FOR SEQ ID NO:77:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:77:
CTACCTACGA TCTGACTAGC GGCTTTATCC GTAACCTCTT AGTGGGCCGC 50
NCGCTTCACA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:78:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:78:
CTACCTACGA TCTGACTAGC ATGGGAGAAC ACTTAGCCTT CATCCGTTCC 50
TCCTAGTGGG GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:79:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:79:
CTACCTACGA TCTGACTAGC CGCGCGTACG AGCACCTTCA TCCGTCCCTC 50
CTAGTGGGGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:80:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 82 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:80:
CTACCTACGA TCTGACTAGC GGCCGTAASC AAGCCTTYWT CCGTMACCTA 50
CTYAGTGGGG KRGCTTACTC TCATGTAGTT CC 82

(2) INFORMATION FOR SEQ ID NO:81:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE DNA:
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:81:
CTACCTACGA TCTGACTAGC NNNNNNNNCT CTTTCATCCG TACCTCCCAG
TGGAGAACGC GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:82:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:82:
CTACCTACGA TCTGACTAGC ACCGGAGTAC TTCATCCGTC CCTTCTAGTG 50
GGTACCCGTA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:83:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:83:
CTACCTACGA TCTGACTAGC TCCGGAGTAC TTCATCCGTY CCTTCTAGTG 50
GGTACCCGTA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:84:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:84:
CTACCTACGA TCTGACTAGC AGGGATGTTC ATCCGTTCCT CTCAGTGGCA 50
TCCCGTGGCT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:85:
(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:85:

CTACCTACGA TCTGACTAGC NNNNGGTTCA TCCGTTCCTT CTAGTGGCCA 50
CCTGGATGCA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:86:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:86:

CTACCTACGA TCTGACTAGC TGGCATTCAT CCGTCTCTCC TAGTGGTGCC 50
TTGTCCCCCA GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:87:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:87:

CTACCTACGA TCTGACTAGC CCTTCATCCG TTACTCTTAG TGGGGGCTTG 50
CGATTCGAGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:88:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:88:

CTACCTACGA TCTGACTAGC TGCTGGACAA TTGATCCGTT ACTCTTAGTG 50
GTTGTGTGCT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:89:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:89:

CTACCTACGA TCTGACTAGC ACGGGTGAGT TGATCCGTTA CTCTTAGTGG 50
TGAACCTTGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:90:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:90:

CTACCTACGA TCTGACTAGC ACGGGTGAGT TGATCCGTCA CTCTTAGTGG 50
TGAACCTTGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:91:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:91:

CTACCTACGA TCTGACTAGC ACAGGTGAGT TGATCCGTTA CTCTTAGTGG 50
TGAACCTTGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:92:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:92:

CTACCTACGA TCTGACTAGC ACTGGTGAKT TGATCCGTCA CTCTTAGTGG 50
TGAWCCTTGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:93:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 29 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:93:
CACGTTTATC CGTCCCTCCT AGTGGCGTG 29

(2) INFORMATION FOR SEQ ID NO:94:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 37 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:94:
GGGGCACGTT TATCCGTCCC TCCTAGTGGC GTGCCCC 37

(2) INFORMATION FOR SEQ ID NO:95:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:95:
CTACCTACGA TCTGACTAGC GGCGTTTTAC TTACTGGTCT TAGACCGGAG 50
AGACACAGTC GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:96:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 85 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:96:
CTACCTACGA TCTGACTAGC GGCGTTTTAC GTGTACKGGT CATGTGAGRC 50
SGGAGAGACA CRGTKGCTTA CTCTCATGTA GTTCC 85

(2) INFORMATION FOR SEQ ID NO:97:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 81 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:97:
CTACCTACGA TCTGACTAGC GATGGGNGGG GGACGTGCTG ATTTCCCACT 50
TCATATTTCG TGCTTACTCT CATGTAGTTC C 81

(2) INFORMATION FOR SEQ ID NO:98:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:98:
CTACCTACGA TCTGACTAGC GAGGGTGTGG GACGTGCTGA TTTCCCACTT 50
CATATTTCGT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:99:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:99:
CTACCTACGA TCTGACTAGC CTGGGKKTCC CGCGGAKCKG TTACCAGTTA 50
TGGGGCAAGG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:100:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:100:
CTACCTACGA TCTGACTAGC CTGGGGGTSC CGCGGAGCGT GTTACCAGTT 50
ATGGGGCAAT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:101:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:101:
CTACCTACGA TCTGACTAGC CTGGGGGTCC CGCGTAGCGT GTTACCAGTT 50
ATGGGGCAAT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:102:
(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:102:

CTACCTACGA TCTGACTAGC CCTACCGTCG KCGCGGTTAA GGAAACTACB 50
GCCTTTTACB GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:103:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:103:

CTACCTACGA TCTGACTAGC ACCGCTAGTT TCGAGGTTGG ACGTGTTTGC 50
CGTGTCGATT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:104:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:104:

CTACCTACGA TCTGACTAGC ACCGCTAGTT TCGAGGTTGG ACGTGTTTGC 50
CGTGTCGATT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:105:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:105:

CTACCTACGA TCTGACTAGC GTTGGACGGT TACGTTTCCT CATGGCAACC 50
CAGCTAGATC GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:106:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 78 base pairs
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:106:

CTACCTACGA TCTGACTAGC TTTCCCTCGA CGGGTGCCCA CTGCGGCATG 50
GGTTAAGAGC TTACTCTCAT GTAGTTCC 78

(2) INFORMATION FOR SEQ ID NO:107:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:107:

CTACCTACGA TCTGACTAGC TGGGGCAGCT TTGCGNGGGT CCTACGTTTT 50
ACDTTTGCCG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:108:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:108:

CTACCTACGA TCTGACTAGC NNCGTCCTTC CAGTGGTGGA GTACCACCCG 50
TCCGGCACTT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:109:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:109:

CTACCTACGA TCTGACTAGC CGCGGGTGAT CGGATTRCCC TGCATCTCCR 50
CCTGATTCTT GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:110:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:110:
CTACCTACGA TCTGACTAGC GATNGCGTTT CGATACTGCT TCTGCGGAGT 50
CACACAGCTC GCTTACTCTC ATGTAGTTCC 80

(2) INFORMATION FOR SEQ ID NO:111:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 79 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:111:
CTACCTACGA TCTGACTAGC AGGCGGGTNC CCTCTGGCGG AACACTTTGC 50
TGTTGTCCTG CTTACTCTCA TGTAGTTCC 79

(2) INFORMATION FOR SEQ ID NO:112:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 80 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:112:
CTACCTACGA TCTGACTAGC NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN 50
NNNNNNNNNN GCTTACTCTC ATGTAGTTCC 80

CLAIMS:

1. A method of identifying nucleic acid ligands to Immunoglobulin E (IgE), comprising:

a) preparing a candidate mixture of nucleic acids;

b) contacting said candidate mixture of nucleic acids with IgE, wherein nucleic acids having an increased affinity to IgE relative to the candidate mixture may be partitioned from the remainder of the candidate mixture; and

c) partitioning the increased affinity nucleic acids from the remainder of the candidate mixture; and

d) amplifying the increased affinity nucleic acids to yield a mixture of nucleic acids enriched for nucleic acid sequences with relatively higher affinity and specificity for binding to IgE, whereby nucleic acid ligands of IgE may be identified.

2. The method of claim 1 further comprising:

e) repeating steps b), c), and d).

3. The method of claim 1 wherein said candidate mixture of nucleic acids is comprised of single stranded nucleic acids.

4. The method of claim 3 wherein said single stranded nucleic acids are ribonucleic acids.

5. The method of claim 3 wherein said single-stranded nucleic acids are deoxyribonucleic acids.

6. The method of claim 4 wherein said nucleic acids are modified nucleic acids.

7. The method of claim 6 wherein said nucleic acids are 2'-amino (2'-NH_2) modified ribonucleic acids.

8. A nucleic acid ligand to IgE identified according to the method of claim 1.

9. A purified and isolated non-naturally occurring RNA ligand to IgE wherein said ligand is selected from the group consisting of the sequences set forth in Table 1, or the corresponding DNA sequences thereof or the corresponding complementary sequences thereof.

10. The RNA ligand of claim 9 wherein said ligand is selected from the group consisting of SEQ ID NOS:7 to 41 and 43 to 48.

11. A purified and isolated non-naturally occurring RNA ligand to IgE wherein said ligand is substantially homologous to and has substantially the same ability to bind IgE as a ligand selected from the group consisting of the sequences set forth in Table 1, or the corresponding DNA sequences thereof or the corresponding complementary sequences thereof.

12. A purified and isolated non-naturally occurring RNA ligand to IgE wherein said ligand has substantially the same structure and substantially the same ability to bind IgE as a ligand selected from the group consisting of the sequences set forth in Table 1, or the corresponding DNA sequences thereof or the corresponding complementary sequences thereof.

13. A purified and isolated non-naturally occurring RNA ligand to IgE comprising the sequences selected from the group consisting of SEQ ID NOS: 45

and 46, or the corresponding DNA sequences thereof or the corresponding complementary sequences thereof.

14. The ligand of claim 13 comprising the sequence of SEQ ID NO: 45.

15. A purified and isolated non-naturally occurring DNA ligand to IgE wherein said ligand is selected from the group consisting of the sequences set forth in Tables 5 and 6, or the corresponding RNA sequences thereof or the corresponding complementary sequences thereof.

16. The DNA ligand of claim 15 wherein said ligand is selected from the group consisting of SEQ ID NOS:51-111.

17. A purified and isolated non-naturally occurring DNA ligand to IgE wherein said ligand is substantially homologous to and has substantially the same ability to bind IgE as a ligand selected from the group consisting of the sequences set forth in Tables 5 and 6, or the corresponding RNA sequences thereof or the corresponding complementary sequences thereof.

18. A purified and isolated non-naturally occurring DNA ligand to IgE wherein said ligand has substantially the same structure and substantially the same ability to bind IgE as a ligand selected from the group consisting of the sequences set forth in Tables 5 and 6, or the corresponding RNA sequences thereof or the corresponding complementary sequences thereof.

19. A method for inhibiting the function of IgE comprising administering a pharmaceutically effective amount of an IgE nucleic acid ligand.

20. The method of claim 19 wherein said IgE nucleic acid ligand is identified according to the method of claim 1.

21. The method of claim 20 wherein said ligand is selected from one of the ligands of SEQ ID NOS. 7 to 41 and 43 to 48.

22. The method of claim 20 wherein said ligand is selected from one of the ligands of SEQ ID NOS. 51 to 111.

INTERNATIONAL SEARCH REPORT	International application No. PCT/US95/12401

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) :C07H 21/02, 21/04; C12P 19/34; C12Q 1/68
US CL :435/6, 91.2; 536/22.1

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 435/6, 91.2; 536/22.1

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Please See Extra Sheet.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO, A, 92/14843 (GILEAD SCIENCES, INC.) 03 September 1992, pages 29-30, 38, 59, and 136.	1-22
Y	Autoimmunity, Volume 8, Number 1, issued 1990, S. Nagpal et al., "Demonstration of IgE Antibodies to Nucleic Acid Antigens in Patients with SLE", pages 59-64, see abstract.	1-22

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
15 DECEMBER 1995	17 JAN 1996
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231	Authorized officer STEPHANIE W. ZITOMER, PH.D.
Facsimile No. (703) 305-3230	Telephone No. 703-308-0196

B. FIELDS SEARCHED
Electronic data bases consulted (Name of data base and where practicable terms used):

APS, DIALOG: FILES: BIOSIS PREVIEWS; DISSERTATION ABSTRACTS ONLINE; DERWEN BIOTECH. ABS; CURRENT BIOTECH. ABS, ROYAL SOCIETY OF CHEMISTRY; INPADOC; DERWENT WORLD PATENTS; PASCAL; LIFE SCIENCES COLL.; MEDLINE; EMBASE; CA SEARCH. SEARCH TERMS: IMMUNOGLOBULIN, E, NUCLEIC, APTAMER, IGE